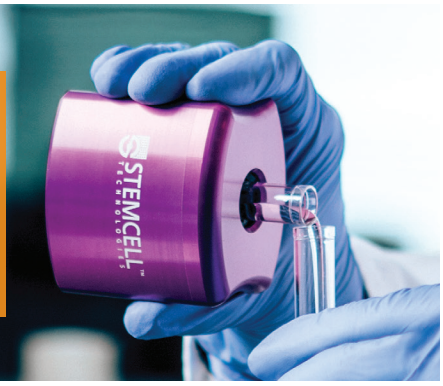
A SMARTER WAY TO ISOLATE CELLS

Cell Separation with EasySep™



Developed with Efficiency in Mind

Working efficiently is key to overcoming the demands of scientific research. That's why our scientists developed EasySep™—a smarter, more efficient way to isolate cells.

At STEMCELL Technologies we strive to provide you with the necessary information to make the right decisions for your research. Here are the facts about EasySep™, backed by data, so you can see the EasySep™ advantages for yourself.

How EasySep™ Works

EasySep™ is a column-free immunomagnetic system for the fast and easy isolation of cells that are immediately ready for downstream applications. Cells of interest are targeted with antibody complexes and magnetic particles for negative or positive selection using an EasySep™ magnet (Figure 1). In as little as 8 minutes (Figure 2), you can obtain highly purified cells from a variety of sample sources, including peripheral blood mononuclear cells, whole blood, leukapheresis products, bone marrow, and cord blood.

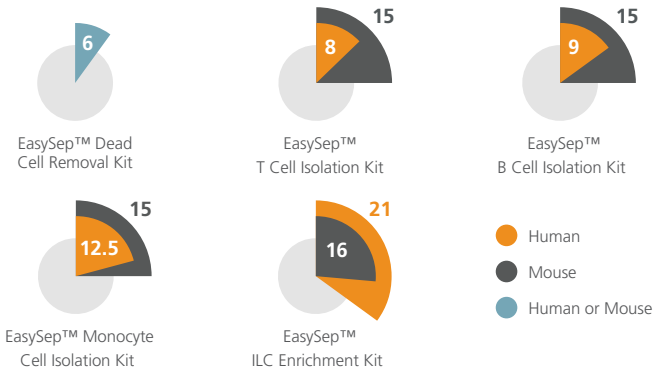


Figure 2. Cell Isolation Protocol Lengths

Time taken (in minutes) to isolate cells using select EasySep™ kits.

> "[EasySep™] enables us to isolate B cells from the spleen really fast with good viability. That is what we need. You don't have to stand in front of a column and wait for it to drip. The viability of the cells that you recover tends to be higher just because it is so fast."
>
> Steevenson N., PhD
> National Institutes of Health

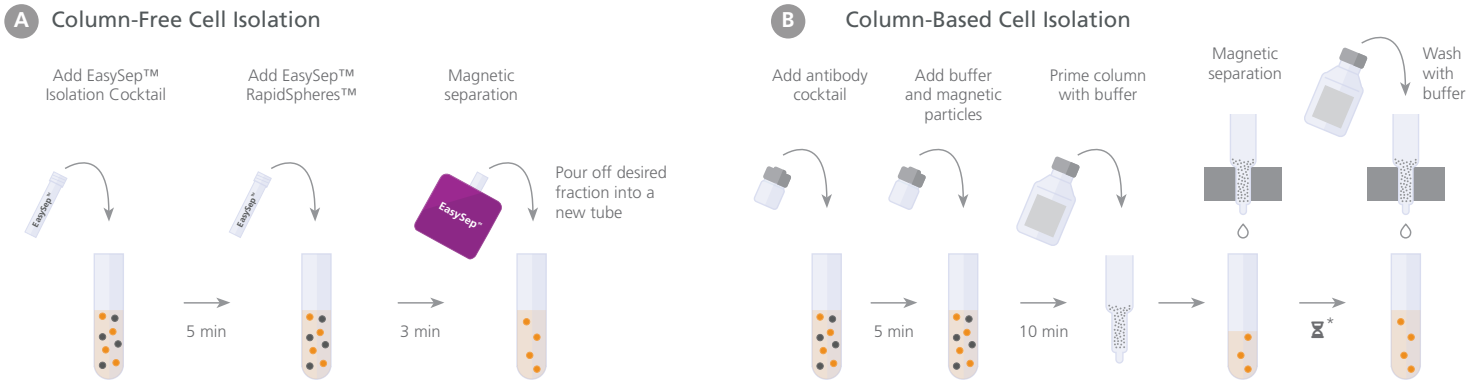


Figure 1. Comparison of Two Different Immunomagnetic Cell Isolation Protocols

Steps involved in the isolation of human T cells by negative selection using two different immunomagnetic cell isolation technologies are shown: (A) column-free EasySep™ technology (time taken may vary depending on the type of magnet used) and (B) a competitor's column-based technology (*elution time varies).

Benefits of an Efficient Workflow

Adopting an efficient cell isolation technology like EasySep™ as the first step in your experimental workflow ensures that your research has a strong foundation.

- Fast protocols preserve cell quality and allow you to get to your downstream applications sooner.
- Robust and optimized protocols minimize variability between samples and allow you to isolate a large number of samples in a short period of time.

Maintain High Cell Viability

Column-free EasySep™ technology allows you to get to your downstream experiments quickly, without extended manipulation of your sample. With the simple, stress-free EasySep™ protocol, cells remain highly viable after separation (Figure 3).

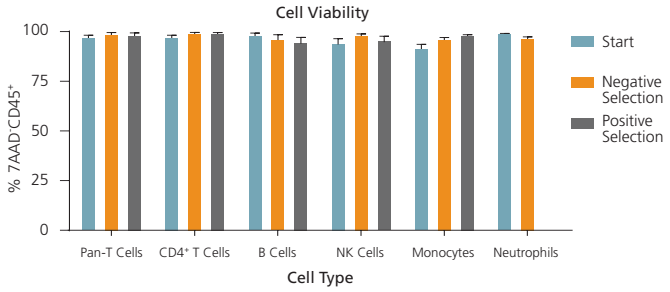


Figure 3. Cells Isolated Using EasySep™ Show Comparable Viability to Starting Samples

Immune cells were isolated from processed leukapheresis or peripheral blood samples using EasySep™ positive selection or negative selection kits. Pre- and post-isolation samples were stained with the cell viability dye 7-AAD and appropriate cell surface markers, and were assessed by flow cytometry. Cells isolated using EasySep™ showed no significant decrease in viability compared to the starting samples. Data shown as mean ± SEM; n = 3 - 7.

Minimize Protocol Optimization

Our scientists have carefully optimized EasySep™ protocols and reagents so you don't have to.

- Cocktails are titrated to ensure proper cell labeling and to avoid epitope blocking (Figure 4).
- Magnetic particles are titrated to minimize non-specific binding.
- Protocols are designed to ensure robust, optimal performance across samples.

> "EasySep™ allows me to pre-enrich my samples for FACS much faster and with less handling. Monocytes are so plastic and sensitive to external factors, and speed is essential when working with these cells. EasySep™ is the fastest separation kit out there."
>
> Sara M., PhD
> University of Queensland

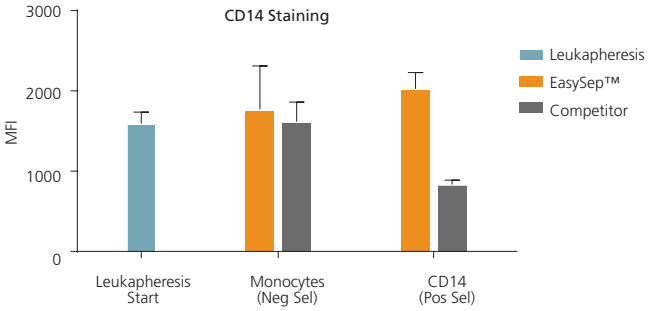
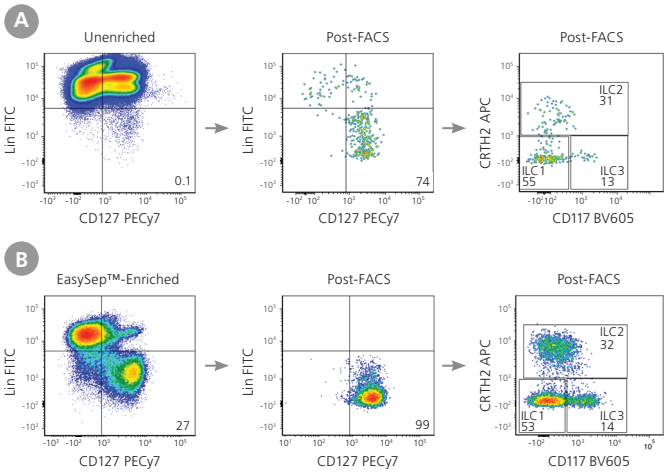


Figure 4. CD14 Epitopes Are Not Blocked Following Cell Isolation Using EasySep™

Cells were isolated by negative selection (Neg Sel) or positive selection (Pos Sel) methods using EasySep™ or a competitor's column-based technology. Isolated cells were stained using an anti-CD14 antibody (clone M5E2), and assessed by flow cytometry. EasySep™-isolated cells showed similar levels of CD14 staining (MFI) compared to unprocessed CD14⁺ cells (Leukapheresis Start). Data shown as mean ± SEM; n = 4 - 5.

Reduce or Eliminate Cell Sorting Time

By using EasySep™ as a pre-enrichment step when isolating rare or complex cell types, you can significantly reduce or eliminate the time you spend performing fluorescence-activated cell sorting (FACS; Figure 5) while maintaining the original subset ratios.



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	START	PRE-FACS	POST-FACS		TIME (MIN)	
	# Starting Cells	Purity	Purity	# Total ILCs	Sample Prep Time	FACS Time
Unenriched	2 x 10^9	0.1%	74%	6.9 x 10^5	15 min	3200 min (53 h)*
EasySep™-Enriched	2 x 10^9	27%	99%	3.7 x 10^5	30 min	12 min

*Time was extrapolated

Figure 5. Pre-Enrichment With EasySep™ Significantly Reduces the Time Required to Obtain Purified ILCs by FACS

Starting with a fresh leukapheresis sample, human innate lymphoid cells (ILCs) were isolated by fluorescence-activated cell sorting (FACS) in parallel from an unenriched or an EasySep™-enriched sample. (A) In an unenriched sample, ILC frequency was assessed by flow cytometry at the start and after one round of FACS. (B) In an EasySep™-enriched sample, ILC frequency was assessed immediately after EasySep™ enrichment, and again after one round of FACS. (C) Corresponding purities and FACS times at each stage are reported.

Efficiency Without Compromising Functionality

EasySep™ kits are designed with cell functionality in mind. During product development, our scientists characterize isolated cells using a variety of relevant functional assays to ensure that isolated cells are representative of their original state.

Isolated Cells Are Responsive to Stimuli

Cells isolated using EasySep™ are functional and respond appropriately to a variety of stimuli (Figures 6 and 7).

- Isolated cells remain unactivated in the absence of stimulation.
- Stimulated cells show an activated phenotype and produce appropriate cytokines.

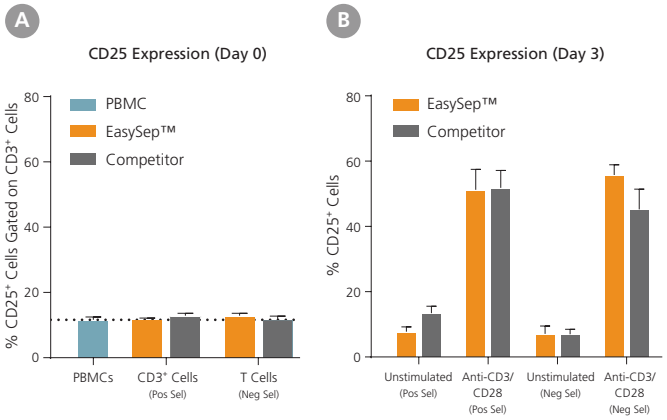


Figure 6. T Cells Isolated Using EasySep™ Show an Appropriate Activation Phenotype

Human T cells were isolated from peripheral blood mononuclear cells (PBMCs) by negative selection (Neg Sel) or positive selection (Pos Sel) using EasySep™ or a competitor's column-based system. Isolated T cells were assessed for CD25 expression immediately after isolation (Day 0) and after 3 days in culture with or without anti-CD3/CD28 stimulation. (A) At Day 0, isolated T cells expressed similar levels of CD25 compared to CD3+ cells in unmanipulated PBMCs. (B) At Day 3, cells remained unactivated in the absence of stimulation, while stimulated cells expressed the activation marker CD25. Data shown as mean ± SEM; n = 3.

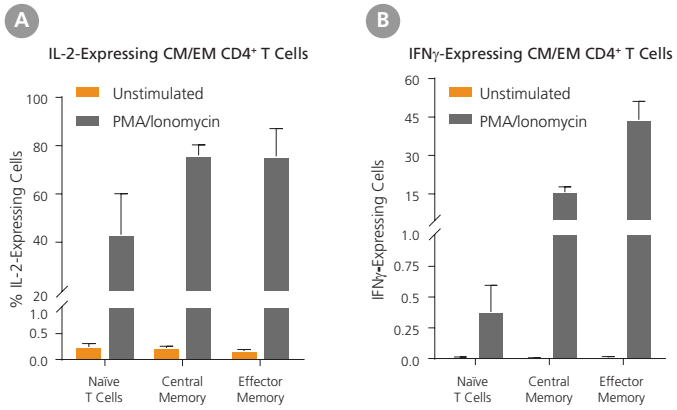
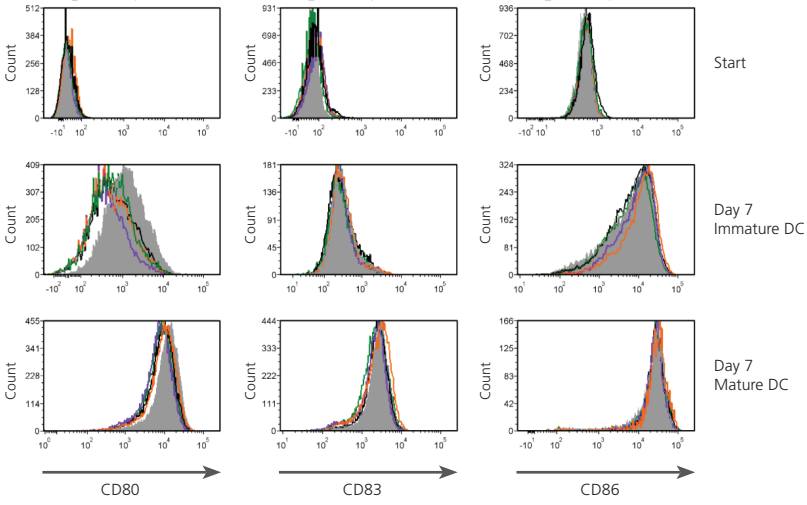


Figure 7. Central Memory and Effector Memory CD4+ T Cells Isolated by EasySep™ Produce Cytokines When Stimulated

T cell subsets were isolated using the EasySep™ Human Central and Effector Memory CD4+ T Cell Isolation Kit. Isolated cells were cultured in medium with or without PMA and ionomycin stimulation for 4 hours. T cell subsets were assessed for the expression of IL-2 (A) and IFNγ (B) by intracellular flow cytometry. In the absence of stimulation, T cell subsets produced low levels of IL-2 and IFNγ. (A) When stimulated, central memory (CM) and effector memory (EM) CD4+ T cell subsets showed significantly higher IL-2 expression compared to naïve T cells. (B) EM CD4+ T cells expressed higher levels of IFNγ than CM CD4+ T cells when stimulated. Both CM and EM CD4+ T cell subsets showed higher IFNγ expression than naïve T cells. Data shown as mean ± SEM; n = 3.

Isolated Cells Differentiate and Mature As Expected

EasySep™-isolated cells express appropriate surface markers following differentiation and maturation (Figure 8).



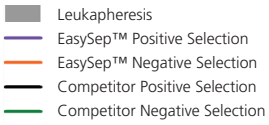


Figure 8. Monocytes Isolated Using EasySep™ Differentiate and Mature Appropriately Upon Stimulation

Human monocytes were isolated using EasySep™ or competitor products and then cultured and differentiated into mature dendritic cells (DCs). On Day 0, cells from a leukapheresis sample were plated and monocytes were adherence-selected for 2 hours. Non-adherent cells were washed away and adherent cells were cultured for 7 days. On Day 5, cells were cultured with maturation supplement for 2 days (mature DCs) or without maturation supplement (immature DCs). The expression of CD80, CD83, and CD86 in immature and mature DCs was determined by flow cytometry. At Day 7, cells expressed the mature DC markers CD80, CD83, and CD86.

Efficiency Without Compromising Performance

High Purity and Recovery

Optimized protocols allow for the efficient isolation of both human (Figure 9) and mouse (Figure 10) target cells with high purity and recovery.

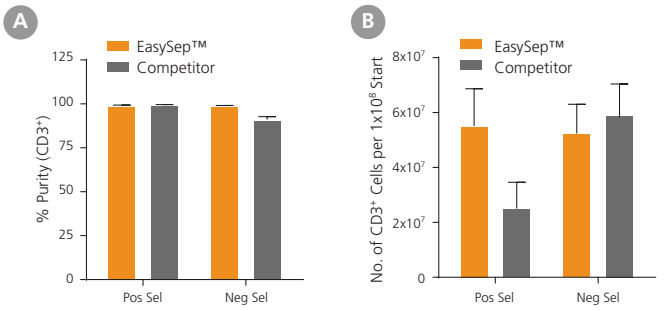


Figure 9. EasySep™ Yields Equivalent or Better Purity and Recovery of Human T Cells Compared to a Column-Based Technology

T cells were isolated by negative selection (Neg Sel) or positive selection (Pos Sel) using EasySep™ or a competitor's column-based technology. EasySep™ isolation yielded comparable or better purity (A) and recovery (B) to the competitor's system. Data shown as mean ± SEM; n = 3.

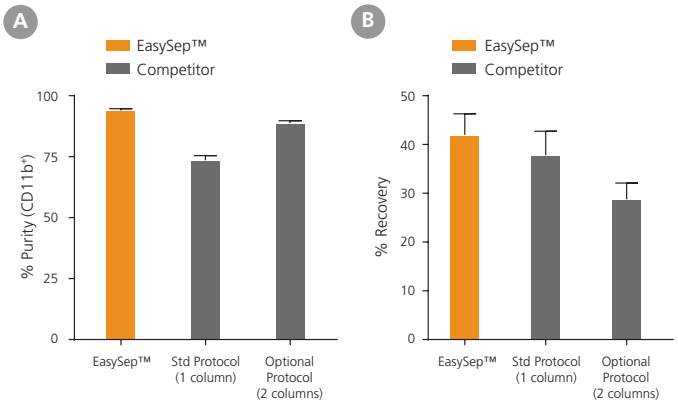


Figure 10. EasySep™ Yields Equivalent or Better Purities and Recoveries of Mouse CD11b$^+$ Cells Compared to a Column-Based Technology

CD11b$^+$ cells were isolated from mouse spleen by positive selection using EasySep™ or a competitor's column-based kit. The competitor's protocol was followed using their standard protocol or their optional "high purity" protocol, which recommended an additional column. EasySep™ isolation yielded comparable or better purity (A) and recovery (B). Data shown as mean ± SEM; n = 4 - 6.

Why Use EasySep™ to Isolate Cells?

FAST AND EASY. Isolate cells in as little as 8 minutes with a simple pour.

HIGH PURITY. Achieve up to 99% cell purity with high recoveries.

GENTLE. Obtain viable, functional cells without the need for columns and washes.

VERSATILE. Isolate cells from various sample sources, including whole blood and leukapheresis samples.

PROVEN. Tested and proven with over 7,000 peer-reviewed publications.

"EasySep™ has allowed us to enrich for rare populations of cells ...[and] allowed us to accurately and efficiently phenotype our cells of interest. EasySep™ helped our lab analyze rare polyclonal antigen-specific CD4$^+$ T and B cell responses in an accurate and cost-effective manner."

Jessica Y., PhD Candidate
University of Minnesota

"We can quickly isolate T cells with high purity and great viability in order to culture them in vitro or do further analysis, like chromatin IP [immunoprecipitation] or RNA sequencing. We have great purities, so EasySep™ works great for these isolations and analyses."

Kyle B., PhD Candidate
University of British Columbia

A Smarter Way to Isolate Cells

We want you to drive science forward, which is why we are committed to developing high-quality, efficient products for your research. Isolate your cells in a smarter, more efficient way with EasySep™. Try it in your own lab and see the advantage yourself.

www.EasySep.com